**United States Department of Agriculture**

Animal and Plant Health Inspection Service

Biotechnology Regulatory Services

4700 River Road
Riverdale, MD
20737

Dr. Robert Stupar, Associate Professor
University of Minnesota
1991 Upper Buford Circle
411 Borlaug Hall
St. Paul, MN 55018

Re: Confirmation of the regulatory status of CRISPR/Cas9 mutagenized *Glycine max* lines 673-7-8 and 673-7-12

Dear Dr. Stupar,

Thank you for your letter received on March 18, 2019, inquiring whether the soybean (*Glycine max*) products described in your letter are regulated articles under 7 CFR part 340. Your letter describes two *G. max* lines developed through CRISPR/Cas9-mediated genome editing to induce a double-stranded break at the target sites, resulting in frame-shift deletions that deactivated the targeted genes, resulting in the intended changes in seed composition traits.

The Plant Protection Act (PPA) of 2000 gives USDA the authority to oversee the detection, control, eradication, suppression, prevention, or retardation of the spread of plant pests or noxious weeds to protect the agriculture, environment, and economy of the United States.

USDA regulates the importation, interstate movement and environmental release (field testing) of certain genetically engineered (GE) organisms that are, or have the potential to be, plant pests. Regulations for GE organisms that are or have the potential to be plant pests, under the PPA, are codified at 7 CFR part 340, "Introduction of Organisms and Products Altered or Produced Through Genetic Engineering Which Are Plant Pests or Which There Is Reason To Believe Are Plant Pests." Under the provisions of these regulations, a GE organism is deemed a regulated article if it has been genetically engineered using a donor organism, recipient organism, or vector or vector agent that is listed in §340.2 and meets the definition of a plant pest, or that is an unclassified organism and/or an organism whose classification is unknown, or if the Administrator determines that the GE organism is a plant pest or has reason to believe it is a plant pest.

In your March 18, 2019 letter, you stated that you transformed *Glycine max* cultivar 'Bert' using the RNA-guided DNA endonuclease enzyme Cas9 to mutate the genes of interest. The nuclease caused double-stranded breaks at the target sites that were

repaired by the plant's DNA repair mechanisms and resulted in frame shift mutations that deactivated the target genes. You stated that the transformation was carried out on your soybean variety using disarmed *Agrobacterium rhizogenes* and that plant pest sequences were used in the plasmid as regulatory elements. You further stated in your letter that you obtained GE line 673-7 which contained a transgenic sequence on chromosome 1. By self-pollinating the originally transformed GE line and screening the parent and progeny lines, you selected non-GE progeny lines 673-7-8 and 673-7-12. Your letter also stated that you used whole genome sequencing to confirm that progeny lines 673-7-8 and 673-7-12 contain no transgenic material nor any plant pest sequences that were used during the transformation.

Based on the information you provided in your letter, USDA has determined that the *Glycine max* progeny lines 673-7-8 and 673-7-12 are not plant pests. The genome edited *G. max* lines do not contain any of the genetic material that was inserted in the GE parent plants for CRISPR/Cas9 editing. The only genetic changes in the genome-edited soybean lines are frame-shift deletions. Since no DNA repair template was provided, the resulting deletions were produced by the plant's own naturally-occurring DNA repair mechanism. Therefore, consistent with previous responses to similar letters of inquiry, APHIS does not consider the genome-edited *Glycine max* lines described in your March 15, 2019 letter to be regulated pursuant to 7 CFR part 340.

USDA is also authorized to protect American agriculture from damage caused by noxious weeds. If USDA determines that a GE plant or introgression of the GE trait into its sexually compatible wild relatives poses a noxious weed risk, USDA would consider regulating the plant under the noxious weed regulation, 7 CFR part 360. USDA has the option to regulate plants under 7 CFR part 360 regardless of whether or not they meet the definition of a regulated article under 7 CFR part 340. *Glycine max* is not listed as a Federal noxious weed pursuant to 7 CFR part 360, and APHIS has no reason to believe that the seed composition phenotype resulting from the genome edits described in your letter would increase the weediness of *Glycine max* or its sexually compatible wild relatives.

Please be advised that the importation of 673-7-8 and 673-7-12 seeds or plants, like all other *Glycine max*, will be subject to Plant Protection and Quarantine (PPQ), permit and/or quarantine requirements. For further information, should you plan to import these *Glycine max* seeds or plants, you may contact the PPQ general number for such inquiries at (877) 770-5990.

Please be advised that your 673-7-8 and 673-7-12 lines of *Glycine max*, while not regulated by APHIS under 7 CFR part 340 may still be subject to other regulatory authorities such as FDA or EPA.

Should you become aware at any time of any issues that may affect the Agency's conclusion regarding this inquiry, you must immediately notify the Agency in writing

of the nature of the issue. We hope that you appreciate our commitment to plant health and support for the responsible stewardship for the introduction of GE plants.

Sincerely,

06/17/2019

Michael J. Firko, Ph.D.
APHIS Deputy Administrator
Biotechnology Regulatory Services
Animal and Plant Health Inspection Service
U.S. Department of Agriculture

Date